Anatomy and Mechanisms of Neurotensin-Dopamine Interactions in the Central Nervous System

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Neurotensin (NT), a tridecapeptide,[1] is widely distributed in the brain of various species and is recognized as a putative peptide neurotransmitter in the mammalian brain. There now exists a growing body of anatomical, neurochemical, electrophysiological, behavioral, and pharmacological evidence that NT interacts with brain dopamine (DA) systems.

NEUROCHEMISTRY

The modulatory influence of NT on brain DA neuronal function is widespread with effects reported for retinal to hypothalamic[2] DA neurons. Light microscopic autoradiographic analyses have shown a dense localization of NT receptors on DA-containing neurons in the substantia nigra zona compacta and the ventral tegmental area (VTA).[3] A facilitatory effect of NT on [^{3}H]-DA release from rat mesencephalic cells in primary culture represents a functional corollary of this receptor distribution.[4] Several studies using 6-hydroxydopamine-induced lesions of the nigrostriatal or mesolimbic DA system in the rat have also demonstrated the presence of NT receptors on presynaptic dopaminergic innervation.[5–8] Along with the dopaminergic terminals, a large percentage of NT receptors is lost in the caudate putamen, nucleus accumbens, and olfactory tubercle following 6-hydroxydopamine lesion of the median forebrain bundle.[8] As well as receptors for both NT and DA existing in the same anatomical location, the neurotransmitters themselves are colocalized within neurons originating from the ventral tegmental area and projecting to either the nucleus accumbens[9] or the prefrontal cortex[10] in the rat. Whether an interaction would have the same physiological relevance in man is unclear because, in contrast to the rat, NT and DA in man are not colocalized in neurons projecting to the prefrontal cortex. The presence of NT receptors on presynaptic dopaminergic terminals in the caudate-putamen correlates well with demonstrated biochemical effects of NT on this pathway. It has been shown that NT modifies DA metabolism in the striatum[11] and facilitates the *in vitro* release of DA from rat striatal slices.[12]

A modulatory interaction between NT and DA receptors on receptor function is also supported by accumulating evidence. *In vitro* findings demonstrate that NT reduces the affinity of [^{3}H]-DA binding sites in membranes from the subcortical

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limbic forebrain and striatum.[13] It has been shown, both *in vivo* and *in vitro,* that exogenous NT decreases the affinity of D2 and D3 DA receptor agonist binding.[14–16] Both D2 and D3 DA receptors are thought to be coupled to G-proteins; however, the effect of NT on the affinity of DA receptors would seem to be via an as-yet-unknown, G-protein-independent mechanism.[14,15] Data indicates that DA receptor activation produces a significant reduction in affinity (increased K_d), and a significant increase in the number of [^{3}H]-NT binding sites (increased B_{max}) in subcortical limbic membranes.[17] Furthermore, rat striatal membranes from the 6-hydroxydopamine-lesioned hemisphere exhibit a significantly enhanced effect of DA (10 nM) on the K_d for [^{3}H]-NT binding sites compared to membranes from the intact side.[6] These data support an inhibitory effect of DA on NT neurotransmission, perhaps mediated by an intramembrane feedback loop of receptor-receptor interactions. Chronic treatment with the neuroleptic haloperidol enhances NT receptor binding in the substantia nigra of the human (postmortem tissue from patients with schizophrenia) and rat brain.[18] Chronic DA receptor blockade leads to a significant increase in NT receptor density in both species. The predicted increase in the number of DA receptors (particularly the D2 subtype) was also seen. Interestingly, electron microscopic analysis revealed an increased number of DA terminals in animals treated with neuroleptics that positively correlated with the cellular increase in the number of NT receptors.[19] These results give further support to the idea that heteroregulatory NT receptor–DA receptor interactions exist within dopaminergic neurons. The increased density of NT receptors following treatment with neuroleptics could lead to increased excitability of DA cells as a compensatory response to the effects of DA receptor blockade.

BEHAVIOR

NT produces a large number of distinct physiological and behavioral effects after systemic and central administration. As examples, the systemic administration of NT produces hypotension, hyperglycemia, smooth muscle contraction, and inhibition of gastric acid secretion.[20] In contrast, after intracerebroventricular (i.c.v.) administration, NT potentiates barbiturate-induced sedation[21] and produces hypothermia,[22] muscle relaxation,[23] antinociception,[24] a reduced food consumption,[25] catalepsy,[26] alterations in locomotor activity[27] and increased serum corticosterone.[2] None of these effects is seen after intravenous NT, indicating that they are centrally mediated. Many of the central effects of NT mimic those seen after administration of neuroleptics, and considerable data now exists to support the hypothesis that NT possesses a pharmacological profile similar to that of neuroleptics. It has been proposed that NT may be an endogenous neuroleptic-like peptide. Differences in the central effects of NT and neuroleptics do exist. For instance, an increase in serum corticosterone levels was not seen following treatment with neuroleptics and although NT does have a stimulatory action on tuberoinfundibular DA neurons, the increase in serum corticosterone does not seem to be mediated via a dopaminergic mechanism.[2] The dose of NT needed to activate DA neurons was much greater than the lowest dose producing a significant increase in serum corticosterone levels, an effect that was not blocked with haloperidol.[2]

Like systemically administered DA antagonists, i.c.v. administration of NT attenuates rat locomotor activity induced by indirect-acting DA agonists.[28] This behavioral interaction of NT with brain DA systems appears to be selective for mesolimbic DA projections. Discrete injection of NT into the nucleus accumbens, like haloperidol, blocked d-amphetamine-induced locomotor behavior;[29] injection of

NT into the caudate-putamen did not antagonize d-amphetamine-induced stereotypic behavior, although haloperidol was potent in this regard. NT has also been shown to antagonize directly the effects of DA in the mesolimbic DA system. NT injection either i.c.v. or directly into the nucleus accumbens produced a dose-dependent reduction in both locomotion and rearing produced by intraaccumbal DA injection.[30] Neurotensin (i.c.v.) has also been shown to block hyperactivity induced by other direct-acting DA receptor agonists such as n-propylnorapomorphine and ADTN.[31,27] These data support the view that NT does not act primarily on presynaptic, but rather on postsynaptic sites of DA neurotransmission in the nucleus accumbens. Further evidence exists to support the multilevel ability of NT to modulate selective brain DA systems. Bilateral injection of NT into the ventral tegmental area produces a dose-dependent increase in locomotion in the rat.[32–34] This effect is consistent with an antagonism by NT of the autoinhibitory effects of DA released from dendrites. Thus, although the behavioral effects of NT, after discrete injection into different brain regions, would seem to be quite different, they can be explained by a common mechanism of interaction with, and antagonism of, DA systems.

ELECTROPHYSIOLOGY

NT has been shown to produce an increase in the *in vitro* firing rate of DA neurons in the zona compacta of the substantia nigra and VTA,[35] while exerting negligible effects on nondopaminergic neurons.[36] *In vitro* studies have demonstrated a linear dose-response curve for the effect of NT on DA cell firing rate,[35,37] although doses exceeding approximately 300 nM produced a decreased firing rate followed by cessation of measurable activity.[35,38] The C-terminal peptide fragment, NT 8–13, possesses similar activity to NT, whereas direct application of the N-terminal fragments, NT 1–8 and NT 1–11, produced no measurable alteration in the firing rate of dopaminergic neurons *in vitro*. Experiments conducted in high-magnesium and low-calcium media suggest that the NT-induced increase in neuronal firing occurs via a calcium-dependent postsynaptic mechanism.[35] Initial data, both *in vitro* and *in vivo,* showed that NT attenuated the DA-induced inhibition of firing in dopaminergic neurons.[39,40] The mechanism of action of this effect remains unknown.

The effects of NT on neuronal firing rate in brain areas outside the mesencephalon are largely unknown. However, a small number of studies show that NT increased the firing rate of individual neurons in various cortical regions.[41,42] The ability of NT to depolarize neurons in slice preparations of medial prefrontal cortex was not affected by pretreatment with phentolamine, propranolol, sulpiride or fluphenazine. *In vitro* electrophysiological studies have found variable effects of NT on hypothalamic neurons. The direct application of NT to preoptic-anterior hypothalamic neurons produced a significant increase in firing rate that persisted in a low-calcium medium;[43,44] however, little or no response was seen when NT was applied to arcuate nucleus neurons.[45] An *in vivo* study has shown a significant inhibitory effect of NT on the firing rate of neurons within the rat thalamus and a variable effect on neurons within the hippocampus.[41]

The electrophysiological effects of acute and chronic treatment with antipsychotic drugs on midbrain dopaminergic neurons have been extensively studied.[46] Most clinically efficacious antipsychotic drugs tested increased the firing rate of dopaminergic neurons in the VTA and zona compacta;[47] repeated administration induced a depolarization inactivation of dopaminergic neurons in the VTA and substantia nigra.[48,49] Bath application of haloperidol produces an attenuation of the

DA-induced inhibition of firing rate in the substantia nigra,[50] whereas no effect was seen after treatment with clozapine.[51]

The available electrophysiological data are in good agreement with neurochemical and behavioral studies regarding an interaction between NT and dopaminergic systems. The actions of NT appear to be independent of other neurotransmitter receptors and the C-terminus of the NT molecule is essential for activity. The electrophysiological data for NT are remarkably similar to those produced by antipsychotic drugs and further support behavioral and electrophysiological data suggesting that NT may be an endogenous antipsychotic.

STRUCTURE AND FUNCTION OF NEUROTENSIN AND DOPAMINE RECEPTORS—INTERACTIONS AT THE LEVEL OF RECEPTOR SIGNAL TRANSDUCTION MECHANISMS

Both the rat and human NT receptor have been molecularly cloned. The cloned rat cDNA encodes a 424-amino-acid peptide,[52] whereas the human cDNA encodes a putative peptide of 418 amino acids with 84% homology to the rat NT receptor.[53] The inferred membrane topology of both the rat and human NT receptor contains seven transmembrane segments, suggesting that the NT receptor represents a member of the superfamily of G-protein-coupled receptors that includes the family of DA receptor subtypes.[54] Recent investigations of the properties of the native NT receptor, as well as NT receptors expressed by both clonal cell lines and in transfected cell systems, have generated a greatly improved understanding of the functional significance and variants of NT receptors. Both autoradiographic and *in situ* hybridization histochemical analyses indicated that brain NT receptors exhibit multiple patterns of ontogenic development that vary greatly between brain regions.[55,56] NT receptor binding sites and mRNA in the rat neocortex demonstrated a transient expression that peaked in the first postnatal week and declined thereafter to adult levels. A second pattern of binding site and mRNA expression indicated an initial expression in late prenatal periods with a gradual development to adult levels within the second week of postnatal life. This pattern was observed in the DA cell body groups of the ventral mesencephalon, the diagonal band, substantia innominata, suprachiasmatic nucleus, and medial habenular nucleus.[56] A third pattern of ontogenic expression was represented by postnatal development of NT receptor binding sites and mRNA with a gradual small decline thereafter. This pattern is apparent in the developing allocortex, tenia tecta, and hippocampus. The fact that the transient expression of neocortical NT receptors occurs long before a neural network is established suggests that NT may play an important role in the development of the neocortical brain structures that precedes its neurotransmitter-like role in the adult brain. A potentially important relevant experiment may involve the use of NT receptor antagonists or immunoinactivation strategies to explore the trophic role of NT in the development of the dopaminergic innervation of the rat neocortex.

The desensitization of the NT receptor response to agonist occupancy by the prolonged exposure to NT receptor agonists has been documented in multiple cell systems and demonstrated for the native receptor. The decrease in receptor number thought to underlie NT receptor desensitization is mediated by a putative mechanism of NT receptor sequestration in response to prolonged agonist exposure. An agonist-induced decrease in cell surface NT receptors has been reported in clonal cell lines[57,58] and in primary cultures of rodent neurons.[59,60] An internalization of rat neostriatal NT receptors and NT followed by their retrograde axonal transport in nigrostriatal DA neurons has been reported in the intact rat brain.[61] These results

support the possibility that NT may be involved in modulating the function of DA neurons at intracellular sites (e.g., gene expression) with long-term actions. Finally, pharmacological studies using either novel NT receptor antagonists (see below) or peptide and pseudo-peptide NT analogs[62] support the concept of a multiplicity of NT receptors. Collectively, the results of these studies support the dissociation of the effects of NT receptor agonists and antagonists on the hypothermic or analgesic effects of NT and the modulatory influences of NT on DA neurons. A subtype of NT receptors apparently mediates the hypothermic and analgesic responses to the peptide, whereas the DA-releasing effects of NT appear to be mediated by a distinct NT receptor subtype that is similar to the cloned high-affinity rat brain NT receptor. An additional level of functional diversity for brain NT systems is provided by the demonstration that the posttranslational processing of the NT/neuromedin N precursor, a 169-residue polypeptide containing one copy each of NT and neuromedin N, is differentially processed in different brain regions.[63]

The regulation by both NT and DA receptors of G-protein-regulated signal transduction systems, as inferred from structural and functional receptor studies, supports the possibility that NT-DA receptor interactions may be expressed in a brain region-specific pattern at the level of post-receptor signaling pathways. Indeed, it was demonstrated in clonal cell lines and transfected cell systems that NT receptor occupancy is associated with alterations in the intracellular content of inositol phosphates,[64] cyclic AMP,[65] cyclic GMP,[66] and calcium.[67] All of these effector systems were shown to be regulated by activation of members of the DA receptor family. Although the NT receptor in these cell lines and systems appears to be pharmacologically similar to the native brain NT receptor, it remains unestablished as to whether the signal transduction mechanisms for the NT receptor in these model cell systems accurately model mechanisms of transmembrane signaling for brain NT receptors. Relatively few studies have investigated these signaling pathways associated with brain NT receptors,[68,69] with no systematic studies having been performed.

Our group has investigated the coupling of rat brain NT receptors with the adenylate cyclase second messenger system and the possible interaction between brain NT and DA receptors at the level of this transduction cascade (TABLE 1). The literature regarding the coupling of the NT receptor with cyclic AMP formation in model cell systems is at the same time both consistent and conflicting. The NT receptor expressed by neuroblastoma N1E115 cells mediates an inhibition of cyclic AMP formation,[70,71] whereas the cloned NT receptor expressed in mammalian cells[72] or Chinese hamster ovary (CHO) cells[73] mediates a stimulation of cAMP formation. The present attempt to characterize the coupling between rat brain NT receptors and cyclic AMP formation examined the effects of NT on cyclic AMP efflux from superfused slice preparations of the rat neostriatum or amygdaloid complex. The selection of a brain slice preparation (i.e., $1.0 \times 0.4 \times 0.4$ mm dimensions) for these studies was based on the observation that neuropeptide receptor-effector coupling has been demonstrated to be dependent on or best demonstrated using intact cell versus homogenate preparations. Also, this preparation, in contrast to cell-free brain homogenates, represents an attempt to replicate for the intact rat brain NT receptor the results of investigations of NT receptor-adenylate cyclase coupling in clonal cell lines and transfected cell systems expressing the NT receptor. The amygdaloid complex was selected for the study of NT receptor coupling with the adenylate cyclase second messenger system because it contains a high concentration of NT as well as NT-immunopositive cells and fibers, with both markers relatively high or dense in the central amygdaloid nucleus.[74] The rat amygdaloid complex also contains a moderate-to-high density of [^{125}I] Tyr_3-NT binding sites.[75]

The effect of NT on the efflux of cyclic AMP from the rat amygdaloid complex

TABLE 1. Effect of Neurotensin on Cyclic AMP Efflux from the Rat Amygdaloid Complex and Caudate-Putamen

Treatment	Cyclic AMP Efflux (% of basal)	
	Amygdaloid Complex	Caudate-Putamen
NT (0.3)	95 ± 6	103 ± 5
NT (3)	84 ± 4	109 ± 7
Forskolin (10)	283 ± 28	—
+ NT (0.3)	221 ± 14[a]	—
Forskolin (10)	322 ± 51	561 ± 66
+ NT (3)	385 ± 40	663 ± 79
PGE_1 (30)	285 ± 15	—
+ NT (3)	331 ± 18	—
Isoproterenol (3)	226 ± 15	—
+ NT (3)	208 ± 18	—
DA (50) + Sulp (30)	—	189 ± 18
+ NT (0.3)	—	173 ± 12
+ NT (3)	—	138 ± 11[a]

NOTE: Values represent determinations ($n = 5$–8) for 5-, 7.5- or 15-min periods of superfusion. Numbers in parentheses represent drug concentrations (μM). Sulp, sulpiride.
[a] $p < 0.05$.

and caudate-putamen was assessed under both basal conditions as well as receptor and non-receptor-mediated increases in cyclic AMP efflux. Superfusion of minced slice preparations of the amygdaloid complex or caudate nucleus with NT-containing buffer did not affect significantly the efflux of cyclic AMP from either region compared to efflux assessed in the absence of NT (TABLE 1). In contrast to the effects of NT on cyclic AMP efflux from neuroblastoma cells expressing NT receptors,[70,71] the forskolin-induced increase in cyclic AMP efflux from the amygdaloid complex or caudate-putamen was not consistently altered by NT. The inhibitory effect of 0.3 but not 3 μM NT on forskolin-stimulated amygdaloid cyclic AMP efflux suggests a coupling of the high-affinity NT receptor with amygdaloid adenylate cyclase. The possible dependence of NT–adenylate cyclase coupling on the interaction with specific receptor types was assessed for prostoglandin E_1 (PGE_1), β-adrenergic receptor, or D1 DA receptor-stimulated cyclic AMP efflux. NT did not affect the PGE_1- or isoproterenol-induced increase in amygdaloid cyclic AMP efflux. NT did, however, produce a concentration-dependent decrease in the D_1 receptor-induced increase in cyclic AMP efflux from the caudate-putamen. NT-DA receptor interactions on cyclic AMP formation were not assessed in the amygdaloid complex as amygdaloid D_1 DA receptors are not coupled to adenylate cyclase. The results obtained in rat brain slice preparations are consistent with a role for effectors other than adenylate cyclase in the transduction of amygdaloid NT receptor signals in the rat brain. Results obtained using experimental controls indicate that these negative findings are not attributable to the degradation of NT during the course of superfusion, an agonist-induced desensitization of NT-receptor function (see above), or the interaction of NT with levocabastine-sensitive binding sites. These results do, however, support an inhibitory influence of the rat striatal NT receptor on transmembrane signaling for the striatal D_1 DA receptor. These latter findings are consistent with a G-protein-mediated effect of NT on the agonist affinity state of the striatal D_1 DA receptor.[76] These results also highlight the conclusion that the generalization of receptor effects from clonal cell lines or transfected cell systems to receptor physiology in the intact central nervous system must await the generation of direct

supporting evidence. Experiments using primary neuronal cultures may be of value in bridging results from model cell systems and brain preparations.

EFFECT OF ANTIPSYCHOTICS ON BRAIN NEUROTENSIN CONTENT AND GENE EXPRESSION

It is increasingly apparent that alterations in DA mechanisms represent insufficient explanations of the pathophysiology or pharmacotherapy of schizophrenia. The similarities between the pharmacological effects of antipsychotics and the central effects of NT (see above) suggest that NT may represent an important element of the final common pathway of the mechanisms of action of antipsychotic drugs. Govoni and co-workers[77] initially reported an increase in NT-like immunoreactivity (NT-LI) in selective areas of the rat brain after treatment with neuroleptics. An increase in NT-LI in the rat nucleus accumbens and striatum was observed following chronic or acute treatment with haloperidol, chlorpromazine, trifluoperazine or pimozide. No drug-induced changes in the concentration of NT-LI in areas of the brain such as the hypothalamus and amygdala were observed. A subsequent preclinical study expanded the investigation of the effects of antipsychotic drugs on brain NT systems by an enhanced anatomical resolution of drug effects and the determination of the comparative effects on NT-LI of a neuroleptic and an atypical antipsychotic agent.[74] The prolonged (14 days) intraperitoneal administration of haloperidol or clozapine did not affect the content of NT-LI in the great majority of the 38 rat brain nuclei or areas examined. However, there was a differential effect of the two antipsychotics in the caudate-putamen, with haloperidol, but not clozapine, increasing the concentration of NT-LI drugs (FIG. 1). Both compounds increased NT-LI in the nucleus

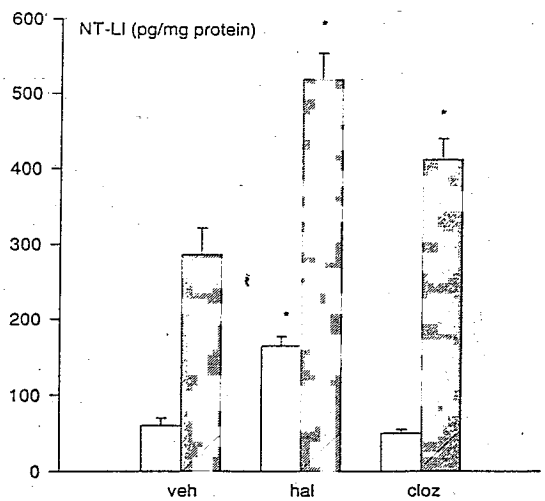

FIGURE 1. Effect of treatment (14 days, i.p.) with 0.3% tartaric acid (veh), haloperidol (hal, 1 mg/kg) or clozapine (cloz, 20 mg/kg) on neurotensin-like immunoreactivity (NT-LI) in the dorsolateral caudate (*open bars*) or the nucleus accumbens (*closed bars*). Results represent the mean (± SEM) of 8 animals per group. $^{*}p < 0.05$ compared to vehicle-treated rats.

accumbens (FIG. 1). The administration of both drugs also produced significant decreases in NT-LI in the medial prefrontal cortex and bed nucleus of the stria terminalis. The brain region- and drug-specific effects of haloperidol and clozapine implicate distinct NT systems in the therapeutic as well as motor side effects of antipsychotic drugs. The findings of a study by Radke and co-workers,[78] consistent with the nontolerating therapeutic effects of haloperidol, showed that long-term (8 months) oral administration of haloperidol produced an enduring increase in NT-LI in the rat nucleus accumbens similar to that observed following 3 weeks of drug administration. Data also indicated that drug-induced alterations in NT concentrations in the rat brain represent a specific response to antipsychotic drugs. Acute (single injection) or repeated treatment (3 weeks) with drugs from other major neuroactive groups, such as tricyclic antidepressants (desipramine), anxiolytics (chlordiazepoxide), and H1 histamine receptor antagonists (diphenhydramine), did not alter rat striatal NT-LI.[79] The drug-induced changes in brain regional NT concentrations also vary in response to different atypical antipsychotics. For example, the atypical antipsychotic drugs sulpiride and rimcazole, in contrast to clozapine, do produce significant increases of NT-LI in the caudate nucleus, but not in the nucleus accumbens.[80] These data further suggest that brain region-specific changes in NT may be related to the distinct therapeutic and side effect profiles of different antipsychotics. Although a direct effect of treatment with antipsychotics on the central NT system has not been demonstrated in man, a normalization of the subnormal cerebrospinal fluid concentrations of NT in a subgroup of drug-free schizophrenics was noted after a course of treatment with antipsychotics.[81] Subsequent clinical studies have largely confirmed and extended these findings.[82]

Although a large amount of accumulated evidence supports brain region–specific and drug dose–dependent changes in NT content in response to antipsychotic drug treatment, the mechanisms underlying these drug effects are not known. Over the past five years, Merchant and co-workers have extensively examined the effects of the typical neuroleptic haloperidol and the atypical antipsychotic clozapine on striatal NT gene expression in the rat brain. A single dose of haloperidol rapidly (within 30 min) increased the expression of NT/N mRNA in the dorsolateral striatum, whereas clozapine produced virtually no effect.[83] The maximal effect of haloperidol was seen at 7 h postinjection, at which time the levels of NT/N mRNA were an order of magnitude higher than those of controls. A small but significant increase in NT/N mRNA was seen in the shell of the nucleus accumbens after treatment with either haloperidol or clozapine.[83] These data support further the possibility that NT neurons in the nucleus accumbens may in part mediate the therapeutic effects common to diverse antipsychotics, whereas NT neurons in the caudate-putamen may be involved in mediating the motor side effects of neuroleptics. Consistent with this theme, Merchant and Dorsa[84] also showed that drug-induced increases in the level of NT/N mRNA within the caudate nucleus were specific to neuroleptic antipsychotics (haloperidol, fluphenazine), whereas increases in the nucleus accumbens occurred after treatment with either neuroleptic or atypical antipsychotics (haloperidol, fluphenazine, clozapine, remoxipride, and thioridazine). Interestingly, they also showed that drug-induced increases in NT/N mRNA expression in the dorsolateral striatum in response to acute treatment with antipsychotics were preceded by a rapid and transient increase in c-fos mRNA.[84] None of the antipsychotics affected c-fos mRNA in the nucleus accumbens. A c-fos antisense oligonucleotide specifically attenuated the haloperidol-induced increase in NT/N mRNA expression, suggesting that an activation of c-fos expression is essential for the effect of haloperidol on NT/N mRNA.[85] These data suggest mechanistic differences in the effects of antipsychotics on discrete NT systems.

Maximal therapeutic benefit of antipsychotics is derived following prolonged drug administration, so an effect of chronic, rather than acute, antipsychotic treatment on NT/N mRNA would be of greater clinical relevance. An initial repeated dosing study showed an increase in NT receptor mRNA in the rat substantia nigra after treatment with haloperidol (2 weeks), but showed no effect after treatment with clozapine.[86] An increase in NT/N mRNA in the rat caudate nucleus has been shown following 28 days of continuous haloperidol administration, but not following chronic clozapine treatment.[87] However, this drug response was only half that caused by acute haloperidol administration. Increases in NT/N mRNA, of similar magnitude to the effect of acute drug treatments, were seen in the nucleus accumbens after chronic treatment with haloperidol or clozapine.[87]

All of the data collected to date investigating the effect of antipsychotics on NT/N mRNA demonstrate a region-specific pattern of effects on NT/N mRNA which discriminates between neuroleptic and atypical antipsychotics. Effects in the limbic sector of the neostriatum would seem more likely to be involved with therapeutic effects of antipsychotics, and NT/N gene induction in the motor striatal regions may indicate cellular events associated with motor side effects of neuroleptics. However, a direct functional connection has not yet been shown between regional changes in NT/N mRNA expression and antipsychotic drug efficacy or side effect profile.

ROLE FOR ENDOGENOUS CENTRAL NEUROTENSIN IN ATTENTIVE FUNCTIONS SENSITIVE TO ANTIPSYCHOTIC DRUGS AND SCHIZOPHRENIA

The lack of selective, potent nonpeptide NT receptor antagonists has delayed the clarification of the neurophysiology and pharmacology of NT. SR48692 (2-[(1-(7-chloro-4-quinolinyl)-5-(2,6-dimethoxyphenyl) pyrazol-3-yl) carbinylamino] tricyclo (3.3.1.13.7) decan-2-carboxylic acid) was recently proposed as the first selective, high-affinity nonpeptide NT receptor antagonist. Initial *in vitro* data showed SR48692 had a high affinity and selectivity for a wide range of mammalian brain NT receptors.[88] SR48692 also had affinity for peripheral NT receptors, inhibiting the increase in blood pressure induced by exogenous NT in the anesthetized guinea-pig and potently antagonizing the tachycardia and inotropic responses induced by NT in the isolated guinea-pig atria.[89] Few experiments have been carried out with SR48692 *in vivo,* in part due to the limited solubility of the compound in physiological solutions. However, SR48692 has been shown to be active *in vivo,* inhibiting the turning behavior in the mouse induced by intrastriatal NT after both i.v. and oral administration.[88] These initial data suggested that SR48692 may be a useful tool for investigating the interactions between endogenous NT and central dopaminergic pathways. In striatal slices the increase in potassium-evoked [^{3}H]-DA release induced by NT was antagonized by SR48692. However, Steinberg and co-workers[90] showed that the increase in DA metabolism in the VTA following intra-VTA microinjection of NT was not antagonized by SR48692. Using *in vivo* voltammetric techniques, they also found no effect of SR48692 on the increased release of DA in the nucleus accumbens evoked by NT injected into the VTA. The lack of effect of SR48692 in these two studies highlights a complexity of NT interactions with brain DA systems and suggests a NT receptor heterogeneity. Further evidence for NT receptor subtypes was provided by the demonstration that SR48692 failed to antagonize NT-induced hypothermia in the rat or mouse.[90]

Our group has an ongoing research interest in latent inhibition (LI) paradigms as

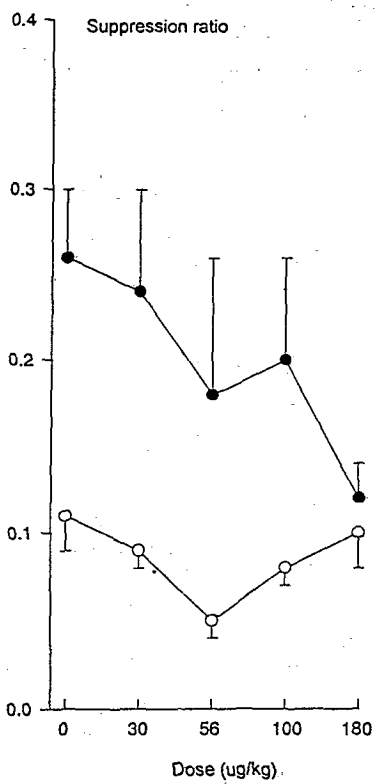

FIGURE 2. Effect of the NT receptor antagonist SR48692 (dose μg/kg) on the latent inhibition (LI) of condition response suppression by stimulus preexposure. Groups of animals receiving 20 preexposures to the to-be-conditioned stimulus (*filled circles*) were compared to non-preexposed rats (*open circles*). Results represent the mean (± SEM) of 6–12 animals per group.

animal behavioral models of stimulus filtering/attentional brain functions sensitive to schizophrenia and targets of the therapeutic actions of antipsychotic drugs.[91] LI represents the negative impact of preexposure to a neutral stimulus on the subsequent ability of that stimulus to serve as a conditioned stimulus. In order to evaluate the antipsychotic potential of endogenous NT, we examined the effects of SR48692 on LI of a conditioned response by stimulus preexposures. We hypothesized that SR48692 would produce an opposite pharmacology to that observed for haloperidol and other antipsychotics. Our initial results (FIG. 2) confirmed this hypothesis by demonstrating a dose-related inhibition of LI by SR48692, in contrast to the increase in suppression ratio seen with haloperidol. These data suggest that NT is recruited in the expression of LI and provide further support for NT as an endogenous antipsychotic.

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